Biochemical and Biophysical Research Communications 425 (2012) 89–93

Contents lists available at SciVerse ScienceDirect

Biochemical and Biophysical Research Communications

journal homepage: www.elsevier.com/locate/ybbrc

Type-I IFN signaling is required for the induction of antigen-specific CD8$^+$ T cell responses by adenovirus vector vaccine in the gut-mucosa

Masaki Shoji [a], Masashi Tachibana [a], Kazufumi Katayama [a], Kyoko Tomita [a], Sayaka Tsuzuki [a], Fuminori Sakurai [a], Kenji Kawabata [b,c], Ken J. Ishii [d,e], Shizuo Akira [f,g], Hiroyuki Mizuguchi [a,b,h,*]

[a] Laboratory of Biochemistry and Molecular Biology, Graduate School of Pharmaceutical Sciences, Osaka University, Osaka, Japan
[b] Laboratory of Stem Cell Regulation, National Institute of Biomedical Innovation, Osaka, Japan
[c] Laboratory of Biomedical Innovation, Graduate School of Pharmaceutical Sciences, Osaka University, Osaka, Japan
[d] World Premier International Immunology Frontier Research Center, Osaka University, Osaka, Japan
[e] Laboratory of Adjuvant Innovation, National Institute of Biomedical Innovation, Osaka, Japan
[f] Laboratory of Host Defense, World Premier International Immunology Frontier Research Center, Osaka University, Osaka, Japan
[g] Research Institute for Microbial Diseases, Osaka University, Osaka, Japan
[h] The Center for Advanced Medical Engineering and Informatics, Osaka University, Osaka, Japan

ARTICLE INFO

Article history:
Received 2 July 2012
Available online 20 July 2012

Keywords:
Innate immunity
Adenovirus vector
Vaccine
Adaptive immunity
Type-I interferon
Gut-mucosa

ABSTRACT

Adenovirus vector (Adv) vaccination at a systemic site, such as intramuscular (i.m.) immunization, can induce antigen-specific CD8$^+$ T cell responses in both systemic and mucosal compartments. It remains unclear, however, how antigen-specific CD8$^+$ T cell response is induced in the mucosa. In this study, we found that type-I IFN signaling is required for the induction of mRNA expression of retinal dehydrogenase in the draining lymph nodes following the i.m. Adv vaccination. We show that type-I IFN signaling is required for the induction of antigen-specific CD8$^+$ T cell response in the gut-mucosal compartment following the i.m. Adv vaccination.

1. Introduction

Replication-incompetent recombinant adenovirus vectors (Adv) that can efficiently transduce exogenous genes are broadly used as gene delivery tools. Because antigen delivery by Adv can induce potent antigen-specific cellular and humoral immune responses in the systemic compartment [1], it is anticipated that Adv vaccine will be effective against infectious pathogens, such as human immunodeficiency virus and influenza virus [2,3]. It was recently reported that the intramuscular (i.m.) immunization of the Adv-expressing simian immunodeficiency virus (SIV)-*gag* induced sustainable and functional SIV-*gag*-specific CD8$^+$ T cell responses in mucosal as well as systemic compartments in mice and rhesus macaques [4–6]. In addition, Ganguly et al. and Kaufman et al. showed that the i.m. Adv vaccine promotes the production of retinoic acid (RA) in dendritic cells (DCs) of draining lymph nodes (DLNs) [6,7]. RA, a vitamin A metabolite, is produced by aldehyde dehydrogenase (Aldh) 1a (retinal dehydrogenase (RALDH)) and is required for imprinting gut-homing capacity on T and B cells [8,9]. Ganguly et al. showed that the RA-dependent upregulation of $\alpha_4\beta_7$ integrin, a gut-homing molecule, on CD8$^+$ T cells induced by Adv requires the activation of nuclear factor-κB (NF-κB) in conventional DCs (cDCs) *in vitro* [6].

Recently, it has been clearly indicated that the innate immune response is essential for effective induction of the adaptive immune response. It is increasingly being considered that the strong induction of antigen-specific CD8$^+$ T cells by Adv vaccine is due to innate immune responses against Adv. In fact, Rhee et al. and others reported that the induction of antigen-specific CD8$^+$ T cell responses by Adv vaccine is significantly reduced in both systemic and mucosal compartments of mice lacking myeloid differentiation protein-88 (MyD88) [10–13]. These results indicate that innate immune signaling is required for the induction of antigen-specific CD8$^+$ T cell responses by Adv vaccine in both systemic and mucosal compartments. However, the antigen-specific immune response was not completely diminished in MyD88-deficient mice [10–13], suggesting that other innate immune signalings are involved in the Adv-mediated antigen-specific CD8$^+$ T cell response in the systemic and mucosal compartments.

The induction of antiviral innate immune response begins with the recognition of viral components by host pattern recognition

* Corresponding author. Address: Laboratory of Biochemistry and Molecular Biology, Graduate School of Pharmaceutical Sciences, Osaka University, 1-6 Yamadaoka, Suita, Osaka 565-0871, Japan. Fax: +81 6 6879 8186.
E-mail address: mizuguch@phs.osaka-u.ac.jp (H. Mizuguchi).

0006-291X/$ - see front matter © 2012 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.bbrc.2012.07.056

receptors (PRRs). The pathogen-recognition by these receptors promotes the maturation of antigen-presenting cells (APCs), and then the activated APCs produce various cytokines which are required to trigger the pathogen-specific adaptive immune response [14–16]. We and others have reported that Adv induces the production of inflammatory cytokines such as IL-6 and IL-12 in cDCs by Toll-like receptor (TLR) 9/MyD88-dependent signaling [17,18]. Zhu et al. showed that the production of type-I IFN by Adv in cDCs is independent of the TLR9/MyD88 signaling, whereas it is dependent on TLR9/MyD88 signaling in plasmacytoid DCs (pDCs) [19]. These observations indicate that Adv can induce innate immune responses through TLR9/MyD88-dependent and – independent signaling. The retinoic acid-inducible gene I (RIG-I)-like receptors (RLRs), including RIG-I and melanoma differentiation-associated gene 5 (Mda5), both of which are expressed in most cell types, are known to recognize cytosolic RNAs [20–22]. RIG-I and Mda5 transduce intracellular signaling through IFN-β promoter stimulator-1 (IPS-1) [23]. This signaling then leads to the production of inflammatory cytokines and type-I IFN. Perreau et al. showed that the levels of type-I IFN positively correlated with titers of Ad5-specific NAbs, suggesting a possibility that type-I IFN signaling controls in the efficacy of Adv vaccine [24]. Thus, it is important to elucidate the effect for adaptive immune responses through the activation of type-I IFN signaling induced by Adv vaccination. We previously reported that IPS-1 is involved in the production of IFN-β in mouse embryonic fibroblasts (MEFs) stimulated by Adv, and Adv-derived virus-associated RNAs (VA-RNAs) which are transcribed by RNA polymerase III are key factors in this process [25]. However, it is unclear whether IPS-1 signaling is involved in the immunogenicity of Adv vaccine.

In this study, we found that IPS-1 and type-I IFN signaling promotes the expression of IFN-β, GM-CSF and RA in the DLNs following the i.m. Adv vaccine. In addition, we found that IPS-1 and type-I IFN signaling are required for the induction of antigen-specific $CD8^+$ T cell responses following i.m. Adv vaccination in the gut-mucosal compartment but not the systemic compartment. These results suggest that type-I IFN signaling is required for the induction of antigen-specific $CD8^+$ T cell responses in the gut-mucosal compartment in an RA-dependent manner following the i.m. Adv vaccine.

2. Materials and methods

2.1. Mice

C57BL/6J (wild-type; WT) mice were purchased from Japan SLC (Hamamatsu, Japan), and IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice (C57BL/6J background) were established as described previously [26]. All mice were housed in an animal facility under a specific-pathogen-free condition and were used at 6–8 weeks of age. All animal experimental procedures used in this study were performed in accordance with the institutional guidelines for animal experiments at Osaka University and the National Institute of Biomedical Innovation.

2.2. Adv production and immunization

The adenovirus type 5 vector-expressing LacZ (Ad-LacZ) was constructed as described previously [27]. Briefly, the expression cassette containing the β-actin promoter with the cytomegalovirus enhancer (CA)-driven [28] LacZ gene was inserted into the E1/E3-deleted adenovirus type 5 genome. This virus was grown in 293 cells by standard techniques. Ad-LacZ was purified with $CsCl_2$ step-gradient ultra-centrifugation, dialyzed with a solution containing 10 mM Tris (pH 7.5), 1 mM $MgCl_2$, and 10% glycerol, and stored in aliquots at −80 °C. Determination of the virus particle (vp) titers was accomplished spectrophotometrically according to the methods of Maizel et al. [29].

All mice were injected under anesthesia in the right and left quadriceps muscles with Ad-LacZ (5×10^9 vp per muscle; total 10^{10} vp per mouse).

2.3. Isolation of mononuclear cells

Peripheral blood mononuclear cells were obtained after lysing red blood cells. Splenocytes were isolated by a standard mechanical disruption procedure, followed by lysis of the red blood cells, and then both components were passed through a nylon mesh. Small bowel lamina propria (LP) lymphocytes were isolated by a standard enzymatic dissociation procedure [30]. In brief, small bowel specimens were removed from Peyer's patches and then cut into small pieces using scissors. These pieces were washed with phosphate-buffered saline (PBS) and then stirred in RPMI 1640 containing 10% fetal calf serum (FCS) and 2 mM EDTA at 37 °C for 20 min to remove intraepithelial cells. The pieces were vigorously shaken, washed five times with PBS to remove EDTA, and then minced and digested using RPMI 1640 containing 10% FCS, collagenase type D (Roche, GmbH, Germany) and DNase I (Roche) at 37 °C for 30 min. Mononuclear cells were then isolated by a discontinuous density gradient procedure (40% and 75%) with Percoll PLUS (GE Helthcare, Tokyo, Japan). The cells that were layered between the 40% and 75% interface were collected as small bowel LP lymphocytes.

2.4. Reverse transcription and quantitative real-time PCR (RT-qPCR)

Total RNA was extracted from inguinal lymph nodes (iLN) lysates using an RNeasy Mini Kit (Qiagen, GmbH, Germany). Total RNA (500 ng) was used to synthesize cDNA using SuperScript VILO (Invitrogen, Carisbad, CA, USA) according to the manufacturer's instructions. The synthesized cDNA was used as a template for real-time PCR, which was performed in triplicate with SYBR Premix Ex Taq™ II (TaKaRa, Shiga, Japan) and each gene-specific primer (*Ifna* forward, 5′-CTTCCACAGGATCACTGTGTACCT-3′; *Ifna* reverse, 5′-TTCTGCTCTGACCACCTCCC-3′; *Ifnb* forward, 5′-CTGGAGCAGCTGAATGGAAAG-3′; *Ifnb* reverse, 5′-CTTCTCCGTCATCTCCATAGGG-3′; *Csf2* forward, 5′-TTTTCCTGGGCATTGTGGTC-3′; *Csf2* reverse, 5′-GGCATGTCATCCAGGAGGTT-3′; *Aldh1a1* forward, 5′-ATGGTTTAGCAGCAGGACTCTTC-3′; *Aldh1a1* reverse, 5′-CCAGACATCTTGAATCCACCGAA-3′; *Aldh1a2* forward, 5′-GACTTGTAGCAGCTGTCTTCACT-3′; *Aldh1a2* reverse, 5′-TCACCCATTTCTCTCCCATTTCC-3′; *Aldh1a3* forward, 5′-GGACAGTCTGGATCAACTGCTAC-3′; *Aldh1a3* reverse, 5′-TCAGGGGTTCTTCTCCTCGAGT-3′; *Gapdh* forward, 5′-CAATG TGTCCGTCGTGGATCT-3′; *Gapdh* reverse, 5′-GTCCTCAGTGTAG CCCAAGATG-3′). PCR and analysis were performed on an Applied Biosystems StepOne Plus Real-time PCR system (Applied Biosystems, Foster City, CA, USA). Relative expression was calculated using the ΔΔCT method, and the mRNA level of each gene was normalized with that of *Gapdh*.

2.5. Tetramer-binding assay

The tetramer-binding assay was performed as previously described [31]. In brief, 1×10^6 lymphocytes from blood, spleen or small bowel LP were incubated with anti-CD16/32 antibody (Ab) (Fc block; eBioscience, San Diego, USA) for 15 min at 4 °C, stained with phycoerythrin (PE)-conjugated H-$2K^b$/β-$gal_{96–103}$ (DAPIYTNV) tetramer reagent (MBL, Nagoya, Japan), and then stained with allophycocyanin (APC)/Cy7-conjugated anti-mouse CD3ε Ab (145-2C11, BioLegend, San Diego, USA) and eFlour™ 450-conjugated anti-mouse CD8α Ab (Ly-2, eBioscience). The stained cells were

analyzed by an LSR II flow cytometer and BD FACSDiva™ Ver 6.1 software (BD Bioscience). Dead cells were excluded by 7-amino-actinomycin D staining (eBioscience).

2.6. Statistics

All results are shown as the mean ± standard error of the mean. Statistical significance was analyzed by the one-way ANOVA among groups. $^{*}p < 0.05$, $^{**}p < 0.01$; compared to WT mice at 8 h. $^{\dagger}p < 0.05$, $^{\dagger\dagger}p < 0.01$; compared to the expression levels of each group at 0 h.

3. Results

3.1. Type-I IFN signaling promotes innate immune responses and the production of RA in draining lymph nodes following i.m. Adv vaccination

It has been clearly shown that the activation of innate immune responses is essential for the effective induction of an adaptive immune response. We previously reported that IPS-1 signaling activated by Adv induces the production of type-I IFNs in MEFs [25]. In addition, it was reported that the iLNs are anatomic sites for the priming and early trafficking of vaccine-induced $CD8^+$ T cells following i.m. administration of Adv vaccine [7]. We speculated that the Adv vaccine imprints the capacity of these $CD8^+$ T cells to migrate into gut-mucosa in iLN through the production of type-I IFN.

To examine this hypothesis, we first analyzed the mRNA expression of *Ifna*, *Ifnb* and *Csf2* (GM-CSF) in the iLN of WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice by RT-qPCR at 8 h after the i.m. administration of Ad-LacZ when the highest expression of *Aldh1a1* in the iLN is induced as shown in a previous report [7]. As the result, we found that the i.m. Ad-LacZ elicited the upregulation of *Ifna* mRNA in WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice (Fig. 1A). The i.m. Ad-LacZ also elicited the upregulation of *Ifnb* and *Csf2* in WT mice, and to a lesser degree in IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice (Fig. 1B and C). It has been reported that RA, a vitamin A metabolite, imprints the gut-homing capacity on T and B cells, and the RA production in DCs was induced by GM-CSF [32] or stimulation through TLR [33]. In addition, DCs express *Aldh1a2* and stromal cells barely express *Aldh1a1* in LNs [34]. Thus, to investigate whether type-I IFN signaling is involved in the promotion of RA production in the iLN, we also analyzed the mRNA expression of *Aldh1a1*, *Aldh1a2* and *Aldh1a3* in the iLN of WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice. We observed the upregulation of *Aldh1a1* but not *Aldh1a2* and *Aldh1a3* in WT mice by the immunization with Ad-LacZ. On the other hand, *Aldh1a1* was not upregulated, and *Aldh1a2 and Aldh1a3* were severely downregulated by the i.m. Ad-LacZ in IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice (Fig. 1D–F). Taken together, these findings suggest that type-I IFN signaling promotes the production of GM-CSF and RA in the iLN following i.m. Adv vaccination.

3.2. Type-I IFN signaling is required for the induction of antigen-specific $CD8^+$ T cell responses by Adv vaccine in the gut-mucosal compartment

Next, we speculated that the downregulation of *Aldh1a* expression might cause the reduction of RA production in IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice, and that the reduced RA production would not sufficiently induce antigen-specific $CD8^+$ T cells in the gut-mucosal compartment following Adv vaccination. To examine this hypothesis, we measured the frequencies of antigen-specific $CD8^+$ T cells in the blood, spleen and small bowel LP of the immunized WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice. Interestingly, the frequencies of β-gal-specific (H-$2K^b$/β-gal_{96-103} (DAPIYTNV) tetramer$^+$) $CD8^+$ T cells in the small bowel LP from IPS$^{-/-}$ mice and IFNAR$2^{-/-}$ mice immunized with Ad-LacZ were significantly reduced as compared with that in WT mice immunized with Ad-LacZ, although the frequencies were equivalent in the blood and

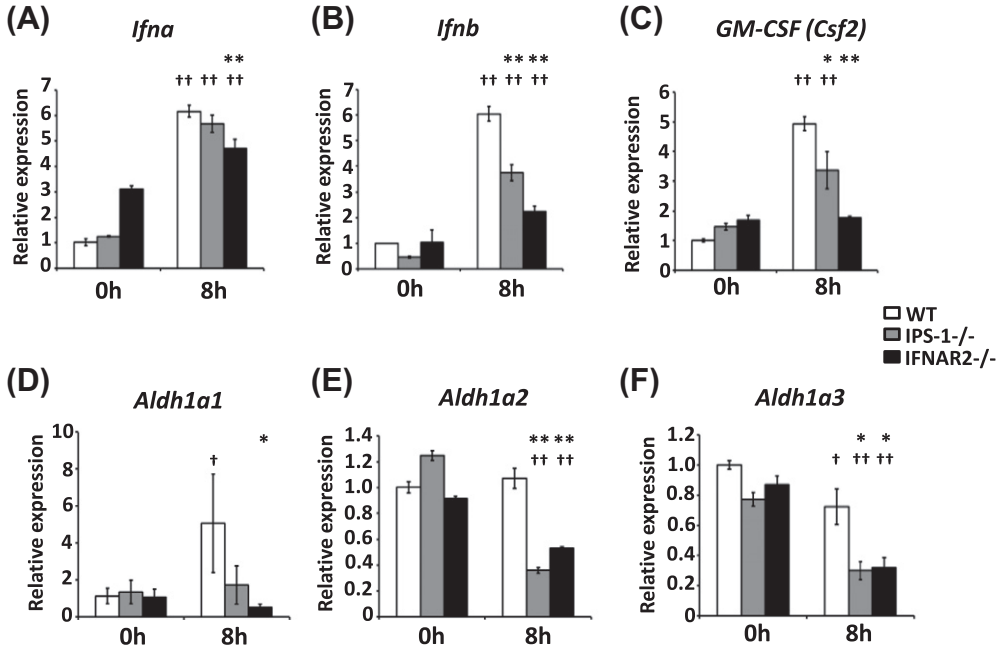

Fig. 1. Relative expression of type-I IFN, GM-CSF and ALDH mRNA in iLN from IPS-$1^{-/-}$ and IFNAR$2^{-/-}$ mice i.m. immunized with Ad-LacZ. At 8 h after the i.m. immunization of 10^{10} vp of Ad-LacZ, total RNA was extracted from whole iLN in WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice. cDNA was synthesized from total RNA, and then expressions of type-I IFN (*Ifna* (A) and *Ifnb* (B)), *GM-CSF* (*Csf2*) (C) and ALDH1A enzymes (*Aldh1a1* (D), *Aldh1a2* (E) and *Aldh1a3* (F)) mRNA were measured by RT-qPCR, normalized by *GAPDH* mRNA. The graphs represent the relative expression of each gene normalized by that of WT mice at the hour 0. Data are shown as the means ± SEM ($n = 3$) and are representative of two independent experiments. $^{*}p < 0.05$, $^{**}p < 0.01$.

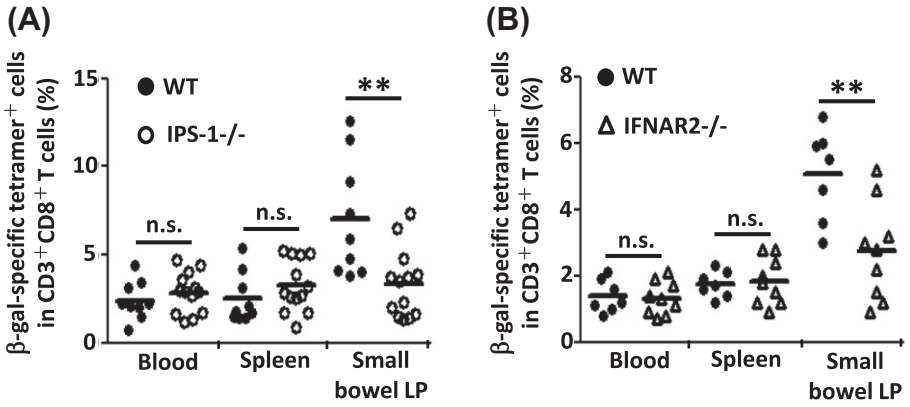

Fig. 2. The frequency of antigen-specific $CD3^+CD8^+$ T cells induced in systemic and gut-mucosal compartments of WT mice, IPS-$1^{-/-}$ mice and IFNAR$2^{-/-}$ mice by the i.m. immunization with Ad-LacZ. At 2 weeks after the i.m. 10^{10} vp of Ad-LacZ immunization, the frequency of β-gal-specific $CD3^+CD8^+$ T cells was measured by H-$2K^b$/β-gal_{96-103} tetramer in the blood, spleen (systemic) and small bowel LP (gut-mucosal) from C57BL/6J wild-type (WT) mice ($n = 9$), and IPS-$1^{-/-}$ mice ($n = 13$) (A), WT mice ($n = 7$) and IFNAR$2^{-/-}$ mice ($n = 9$) (B). Bars indicate averages, and closed circles (WT), open circles (IPS-$1^{-/-}$) or open triangles (IFNAR$2^{-/-}$) indicate individual mouse samples. n.s. indicates not significant. Data pools are representative of three independent experiments. $^{**}p < 0.01$.

spleen among the strains (Fig. 2A and B). Therefore, these results indicate that type-I IFN signaling is required for the induction of antigen-specific $CD8^+$ T cell responses to Adv vaccine in the gut-mucosal compartment but not in the systemic compartment.

4. Discussion

In this study, we showed that type-I IFN signaling activated by the i.m. Adv vaccine promotes the mRNA expression of *Ifnb*, *Csf2 and Aldh1a1* and is required for the induction of antigen-specific $CD8^+$ T cells by Adv vaccine in the gut-mucosal compartment but not the systemic compartment. We observed that the expression of IFN-β, but not IFN-α, after the i.m. immunization of Adv vaccine was prominently affected by IPS-1 or IFNAR2 deficiency. Since the production of IFN-α induced by Adv infection in MEF was undetectable (data not shown), it is speculated that IFN-α would be mainly produced by other type of cells, including DC, and that IFN-β would be produced by fibroblasts such as stromal cells by the i.m. immunization of Adv vaccine. We previously reported that the production of IFN-β in MEF induced by Adv is dependent on IPS-1 signaling and that in DC is partially dependent on IPS-1 signaling [25]. In addition, since previous reports have shown that type-I IFN amplifies its expression through IFNAR [35,36], it is suggested that the expression of IFN-β was severely decreased in IFNAR$2^{-/-}$ mice. Therefore, it is speculated that IPS-1 and type-I IFN signaling contribute to IFN-β, but not IFN-α, production from stromal cells in iLN following the i.m. immunization of Adv vaccine.

It was shown that the stromal cells barely express *Aldh1a1* in LNs [34] and the expression of *Aldh1a1* but not *Aldh1a2* in the iLN of C57BL/6 mice was upregulated following the i.m. immunization of Adv vaccine [7]. From these observations, it is speculated that *Aldh1a1* in the stromal cells of iLN would be induced by Adv vaccination. In support of this hypothesis, we observed the upregulation of *Aldh1a1* in WT mice by the immunization with Adv vaccine. We also observed that the expression of *Aldh1a2* and *Aldh1a3* in the iLN of Adv-immunized WT mice did not change, as was similarly shown for *Aldh1a2* expression in a previous report [7]. On the other hand, Ganguly et al. showed that the i.m. Adv vaccine upregulated the expression of *Aldh1a2* and *Aldh1a3* in cDCs of DLNs [6]. In our study, the expression of *Aldh1a2* and *Aldh1a3* was measured in the whole iLNs consisting of the T and B cells which do not express *Aldh1a2* and *Aldh1a3* and a few cDCs following the i.m. immunization of Adv vaccine. Therefore, we could observe a relatively decreased mRNA expression in cDCs. Taken together, it is suggested that at least the *Aldh1a1* expression contributes to the imprinting of the capacity of antigen-specific CD8+ T cells to migrate into the gut-mucosa.

As shown here, we propose the importance of type-I IFN signaling for the induction of antigen-specific $CD8^+$ T cell responses in the gut-mucosal compartment by Adv vaccine. It is hoped that these findings will contribute to the development of novel vaccines and adjuvants that can enhance the induction of mucosal immune response by immunization through systemic routes.

Acknowledgments

This work was supported by grants from the Ministry of Health, Labour, and Welfare of Japan (H.M.) and The Mochida Memorial Foundation for Medical and Pharmaceutical Research (M.T.).

References

[1] N. Tatsis, H.C. Ertl, Adenoviruses as vaccine vectors, Mol. Ther. 10 (2004) 616–629.
[2] J.Y. Liu, K.L. O'Brien, D.M. Lynch, N.L. Simmons, A. La Porte, A.M. Riggs, P. Abbink, R.T. Coffey, L.E. Grandpre, M.S. Seaman, G. Landucci, D.N. Forthal, D.C. Montefiori, A. Carville, K.G. Mansfield, M.J. Havenga, M.G. Pau, J. Goudsmit, D.H. Barouch, Immune control of an SIV challenge by a T-cell-based vaccine in rhesus monkeys, Nature 457 (2009) 87–91.
[3] M.A. Hoelscher, S. Garg, D.S. Bangari, J.A. Belser, X.H. Lu, I. Stephenson, R.A. Bright, J.M. Katz, S.K. Mittal, S. Sambhara, Development of adenoviral-vector-based pandemic influenza vaccine against antigenically distinct human H5N1 strains in mice, Lancet 367 (2006) 475–481.
[4] D.R. Kaufman, J. Liu, A. Carville, K.G. Mansfield, M.J. Havenga, J. Goudsmit, D.H. Barouch, Trafficking of antigen-specific $CD8^+$ T lymphocytes to mucosal surfaces following intramuscular vaccination, J. Immunol. 181 (2008) 4188–4198.
[5] D.R. Kaufman, M. Bivas-Benita, N.L. Simmons, D. Miller, D.H. Barouch, Route of adenovirus-based HIV-1 vaccine delivery impacts the phenotype and trafficking of vaccine-elicited $CD8^+$ T lymphocytes, J. Virol. 84 (2010) 5986–5996.
[6] S. Ganguly, S. Manicassamy, J. Blackwell, B. Pulendran, R.R. Amara, Adenovirus type 5 induces vitamin A-metabolizing enzymes in dendritic cells and enhances priming of gut-homing CD8 T cells, Mucosal Immunol. 4 (2011) 528–538.
[7] D.R. Kaufman, J. De Calisto, N.L. Simmons, A.N. Cruz, E.J. Villablanca, J.R. Mora, D.H. Barouch, Vitamin A deficiency impairs vaccine-elicited gastrointestinal immunity, J. Immunol. 187 (2011) 1877–1883.
[8] M. Iwata, A. Hirakiyama, Y. Eshima, H. Kagechika, C. Kato, S.Y. Song, Retinoic acid imprints gut-homing specificity on T cells, Immunity 21 (2004) 527–538.
[9] J.R. Mora, M. Iwata, B. Eksteen, S.Y. Song, T. Junt, B. Senman, K.L. Otipoby, A. Yokota, H. Takeuchi, P. Ricciardi-Castagnoli, K. Rajewsky, D.H. Adams, U.H. von Andrian, Generation of gut-homing IgA-secreting B cells by intestinal dendritic cells, Science 314 (2006) 1157–1160.
[10] Z.C. Hartman, A. Kiang, R.S. Everett, D. Serra, X.Y. Yang, T.M. Clay, A. Amalfitano, Adenovirus infection triggers a rapid, MyD88-regulated

transcriptome response critical to acute-phase and adaptive immune responses in vivo, J. Virol. 81 (2007) 1796–1812.
[11] D.M. Appledorn, S. Patial, A. McBride, S. Godbehere, N. Van Rooijen, N. Parameswaran, A. Amalfitano, Adenovirus vector-induced innate inflammatory mediators, MAPK signaling, as well as adaptive immune responses are dependent upon both TLR2 and TLR9 in vivo, J. Immunol. 181 (2008) 2134–2144.
[12] R.W. Lindsay, P.A. Darrah, K.M. Quinn, U. Wille-Reece, L.M. Mattei, A. Iwasaki, S.P. Kasturi, B. Pulendran, J.G. Gall, A.G. Spies, R.A. Seder, $CD8^+$ T cell responses following replication-defective adenovirus serotype 5 immunization are dependent on $CD11c^+$ dendritic cells but show redundancy in their requirement of TLR and nucleotide-binding oligomerization domain-like receptor signaling, J. Immunol. 185 (2010) 1513–1521.
[13] E.G. Rhee, J.N. Blattman, S.P. Kasturi, R.P. Kelley, D.R. Kaufman, D.M. Lynch, A. La Porte, N.L. Simmons, S.L. Clark, B. Pulendran, P.D. Greenberg, D.H. Barouch, Multiple innate immune pathways contribute to the immunogenicity of recombinant adenovirus vaccine vectors, J. Virol. 85 (2011) 315–323.
[14] S. Akira, S. Uematsu, O. Takeuchi, Pathogen recognition and innate immunity, Cell 124 (2006) 783–801.
[15] A. Iwasaki, R. Medzhitov, Regulation of adaptive immunity by the innate immune system, Science 327 (2010) 291–295.
[16] O. Takeuchi, S. Akira, Pattern recognition receptors and inflammation, Cell 140 (2010) 805–820.
[17] E. Basner-Tschakarjan, E. Gaffal, M. O'Keeffe, D. Tormo, A. Limmer, H. Wagner, H. Hochrein, T. Tuting, Adenovirus efficiently transduces plasmacytoid dendritic cells resulting in TLR9-dependent maturation and IFN-alpha production, J. Gene Med. 8 (2006) 1300–1306.
[18] T. Yamaguchi, K. Kawabata, N. Koizumi, F. Sakurai, K. Nakashima, H. Sakurai, T. Sasaki, N. Okada, K. Yamanishi, H. Mizuguchi, Role of MyD88 and TLR9 in the innate immune response elicited by serotype 5 adenoviral vectors, Hum. Gene Ther. 18 (2007) 753–762.
[19] J. Zhu, X. Huang, Y. Yang, Innate immune response to adenoviral vectors is mediated by both Toll-like receptor-dependent and -independent pathways, J. Virol. 81 (2007) 3170–3180.
[20] M. Yoneyama, M. Kikuchi, T. Natsukawa, N. Shinobu, T. Imaizumi, M. Miyagishi, K. Taira, S. Akira, T. Fujita, The RNA helicase RIG-I has an essential function in double-stranded RNA-induced innate antiviral responses, Nat. Immunol. 5 (2004) 730–737.
[21] M. Yoneyama, M. Kikuchi, K. Matsumoto, T. Imaizumi, M. Miyagishi, K. Taira, E. Foy, Y.M. Loo, M. Gale Jr., S. Akira, S. Yonehara, A. Kato, T. Fujita, Shared and unique functions of the DExD/H-box helicases RIG-I, MDA5, and LGP2 in antiviral innate immunity, J. Immunol. 175 (2005) 2851–2858.
[22] J. Andrejeva, K.S. Childs, D.F. Young, T.S. Carlos, N. Stock, S. Goodbourn, R.E. Randall, The V proteins of paramyxoviruses bind the IFN-inducible RNA helicase, mda-5, and inhibit its activation of the IFN-beta promoter, Proc. Natl. Acad. Sci. USA 101 (2004) 17264–17269.
[23] T. Kawai, K. Takahashi, S. Sato, C. Coban, H. Kumar, H. Kato, K.J. Ishii, O. Takeuchi, S. Akira, IPS-1, an adaptor triggering RIG-I- and Mda5-mediated type I interferon induction, Nat. Immunol. 6 (2005) 981–988.
[24] M. Perreau, H.C. Welles, C. Pellaton, B. Gjoksi, L. Potin, R. Martin, A. Harari, A. Bett, D. Casimiro, J. Gall, D.H. Barouch, E.J. Kremer, G. Pantaleo, The number of Toll-like receptor 9-agonist motifs in the adenovirus genome correlates with induction of dendritic cell maturation by adenovirus immune complexes, J. Virol. 86 (2012) 6279–6285.
[25] T. Yamaguchi, K. Kawabata, E. Kouyama, K.J. Ishii, K. Katayama, T. Suzuki, S. Kurachi, F. Sakurai, S. Akira, H. Mizuguchi, Induction of type I interferon by adenovirus-encoded small RNAs, Proc. Natl. Acad. Sci. USA 107 (2010) 17286–17291.
[26] K.J. Ishii, T. Kawagoe, S. Koyama, K. Matsui, H. Kumar, T. Kawai, S. Uematsu, O. Takeuchi, F. Takeshita, C. Coban, S. Akira, TANK-binding kinase-1 delineates innate and adaptive immune responses to DNA vaccines, Nature 451 (2008) 725–729.
[27] H. Mizuguchi, M.A. Kay, T. Hayakawa, In vitro ligation-based cloning of foreign DNAs into the E3 and E1 deletion regions for generation of recombinant adenovirus vectors, Biotechniques 30 (2001). 1112–1114, 1116.
[28] H. Niwa, K. Yamamura, J. Miyazaki, Efficient selection for high-expression transfectants with a novel eukaryotic vector, Gene 108 (1991) 193–199.
[29] J.V. Maizel Jr., D.O. White, M.D. Scharff, The polypeptides of adenovirus. I. Evidence for multiple protein components in the virion and a comparison of types 2, 7A, and 12, Virology 36 (1968) 115–125.
[30] K. Katayama, K. Wada, A. Nakajima, H. Mizuguchi, T. Hayakawa, S. Nakagawa, T. Kadowaki, R. Nagai, Y. Kamisaki, R.S. Blumberg, T. Mayumi, A novel PPAR gamma gene therapy to control inflammation associated with inflammatory bowel disease in a murine model, Gastroenterology 124 (2003) 1315–1324.
[31] M. Shoji, S. Yoshizaki, H. Mizuguchi, K. Okuda, M. Shimada, Immunogenic comparison of chimeric adenovirus 5/35 vector carrying optimized human immunodeficiency virus clade C genes and various promoters, PLoS One 7 (2012) e30302.
[32] A. Yokota, H. Takeuchi, N. Maeda, Y. Ohoka, C. Kato, S.Y. Song, M. Iwata, GM-CSF and IL-4 synergistically trigger dendritic cells to acquire retinoic acid-producing capacity, Int. Immunol. 21 (2009) 361–377.
[33] S. Manicassamy, R. Ravindran, J. Deng, H. Oluoch, T.L. Denning, S.P. Kasturi, K.M. Rosenthal, B.D. Evavold, B. Pulendran, Toll-like receptor 2-dependent induction of vitamin A-metabolizing enzymes in dendritic cells promotes T regulatory responses and inhibits autoimmunity, Nat. Med. 15 (2009) 401–409.
[34] R. Molenaar, M. Greuter, A.P. van der Marel, R. Roozendaal, S.F. Martin, F. Edele, J. Huehn, R. Forster, T. O'Toole, W. Jansen, I.L. Eestermans, G. Kraal, R.E. Mebius, Lymph node stromal cells support dendritic cell-induced gut-homing of T cells, J. Immunol. 183 (2009) 6395–6402.
[35] S. Abraham, A.S. Nagaraj, S. Basak, R. Manjunath, Japanese encephalitis virus utilizes the canonical pathway to activate NF-kappaB but it utilizes the type I interferon pathway to induce major histocompatibility complex class I expression in mouse embryonic fibroblasts, J. Virol. 84 (2010) 5485–5493.
[36] X.D. Shen, B. Ke, H. Ji, F. Gao, M.C. Freitas, W.W. Chang, C. Lee, Y. Zhai, R.W. Busuttil, J.W. Kupiec-Weglinski, Disruption of type-I IFN pathway ameliorates preservation damage in mouse orthotopic liver transplantation via HO-1 dependent mechanism, Am. J. Transplant. 12 (2012) 1730–1739.